

LISTING OF THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the application:

1. (Currently Amended) Electroporation device for the permeabilization of cells (C) contained in a substrate ~~[[(12)]]~~ comprising a signal generator ~~generating means~~ ~~[[(3)]]~~ configured for generating a stimulating signal (S(t)) applied by means of electrodes ~~[[(6,7)]]~~ to the substrate ~~[[(12)]]~~ wherein an electric field (E(t)) permeabilizing the cells membranes is induced; the device ~~being characterized by~~ comprising:

~~[[means]]~~ an element configured for measuring, calculating and monitoring ~~(15,16,23)~~ the instantaneous value of the ~~ratio~~ ratio (GT) of current (ic) flowing between said electrodes ~~[[(6,7)]]~~ and through the substrate ~~[[(12)]]~~ and the voltage (Vp) of the stimulating signal (S(t)) applied to the substrate ~~[[(12)]]~~ by means of said electrodes ~~[[(6,7)]]~~;

said device further comprising a controlling ~~[[means]]~~ ~~(100-170)~~ element configured for applying the stimulating signal in a controlled manner according to the waveform of only an initial portion of the curve C_{GT} representing the ratio (GT) in successive instants after the beginning of the application of the stimulating signal (S(t)).

2. (Currently Amended) Device as claimed in claim 1, wherein said controlling ~~[[means]]~~ ~~(100-170)~~ element comprise a timing ~~means~~ ~~[[(110)]]~~ element configured for applying said stimulating signal for a predetermined period of time T_d and analysing the initial portion of the waveform of curve C_{GT} to detect a minimum value of the curve C_{GT} within the interval $t=0$ and $t=T_d$.

3. (Currently Amended) Device as claimed in claim 1, wherein said controlling ~~[[means]]~~ ~~(100-170)~~ element is configured ~~calculate~~ for calculating the slope of the waveform of curve C_{GT} after ~~[[that]]~~ a minimum in curve C_{GT} has been reached.

4. (Currently Amended) Device as claimed in claim 1, claims, wherein said controlling ~~[[means]]~~ ~~(100-170)~~ element comprise a hazard detecting ~~means~~ ~~[[(120)]]~~ element configured for determining the instantaneous gradient (dG) of said ratio (GT) after a minimum has been reached

in said curve C_{GT} ; said controlling ~~[[means]]~~ element further ~~emprise~~ comprises a first comparing means ~~[[(130)]]~~ element configured for comparing the calculated instantaneous ~~instantaneous~~ gradient dG with at least a reference value (dG_{ref}) and selecting a correcting means ~~(140,145)~~ element for performing an urgent correction to the stimulating signal $S(t)$ in order to avoid lesions, damages or irreversible alterations in said substrate ~~[[(12)]]~~.

5. (Currently Amended) Device as claimed in claim 4, wherein said correcting ~~[[means]]~~ ~~(140,145)~~ element is configured to decrease ~~decrease~~ the voltage of the stimulating signal $S(t)$ in order to prevent deterioration in the cells (C).

6. (Currently Amended) Device as claimed in claim 1, wherein said controlling ~~[[means]]~~ ~~(100-170)~~ element ~~emprise~~ comprises a slope determining means ~~[[(150)]]~~ element configured to calculate ~~calculating~~ the average variation ΔG of said ratio (GT) in a time interval that is successive to the instant T_m wherein a minimum in the curve (C_{GT}) has been reached and that has a pre-determined time width; said controlling ~~[[means]]~~ element further comprising a second comparing means ~~[[(160)]]~~ element configured to compare ~~comparing~~ the calculated average variation ΔG of said ratio (GT) with a reference interval of ΔG values.

7. (Currently Amended) Device as claimed in claim 6, wherein said second comparing means ~~[[(160)]]~~ element performs the following functions: if the calculated average variation ΔG of said ratio (GT) falls within the reference interval ($0 < \Delta G < \Delta G_{obb}$) a continuing means element ~~[[(170)]]~~ are is selected; if the calculated average variation ΔG of said ratio (GT) falls outside the reference interval and it is smaller than both limits delimiting the interval ($\Delta G < 0 < \Delta G_{obb}$) an adjusting element means ~~[[(180)]]~~ are is selected; and if the calculated average variation ΔG of said ratio (GT) falls outside the reference interval and it is greater than both limits delimiting the interval ($\Delta G > \Delta G_{obb} > 0$) a correcting means ~~[[(140)]]~~ element ~~[[are]]~~ is selected.

8. (Currently Amended) Device as claimed in claim 7, wherein said adjusting ~~[[means]]~~ ~~[[(180)]]~~ element is configured to increase the voltage of the stimulating signal in order to increase the value of the electric field $E(t)$ applied to the substrate ~~[[(12)]]~~; said adjusting means

[[180]] element subsequently selecting said ~~means~~ element for calculating and monitoring (15,16,23) the instantaneous value of the said ratio (GT) and said controlling [[means]] element.

9. (Currently Amended) Device as claimed in claim 7, wherein said continuing [[means]] element [[170]] is configured to increase the voltage of the stimulating signal to an objective voltage V_{opt} in order to increase the value of the electric field $E(t)$ applied to the substrate [[12]] so that the value of said average variation ΔG tends to an expected value ΔG_{obb} .

10. (Currently Amended) Device as claimed in claim 1, wherein said controlling ~~means~~ (100-170) element is configured to detect ~~detects~~ [[125]] a minimum of said initial portion of said curve and determining [[126]] the time T_m at which the minimum is reached.

11. (Currently Amended) Device as claimed in claim 10, wherein third comparing ~~means~~ (127) are is provided to compare the detected T_m with threshold values T_{min} and T_{max} ; said third comparing ~~means~~ [[127]] element is configured to perform performing the following operations: if the detected T_m occurs before T_{min} ($T_m < T_{min}$) then a correcting means (140) are is selected; if the detected T_m occurs after T_{max} ($T_m > T_{max}$), then an adjusting means (180) are is selected; and if the detected T_m occurs between T_{min} and T_{max} , then a continuing means (170) are is selected.

12. (Currently Amended) ~~Use of a device as described in claim 1, to extract~~ A method for extracting molecules from [[the]] living cells comprised in [[the]] a substrate, comprising the step of using the device of claim 1 to generate a controlled stimulating signal applied to living cells by means of electrodes while extracting molecules from the living cells.

13. (Currently Amended) ~~Use of a device as described in claim 1, to introduce~~ A method for introducing molecules into living cells comprising the step of using the device of claim 1 to generate a controlled stimulating signal applied to living cells by means of electrodes while introducing said molecules into the living cells.

14. (Currently Amended) ~~Use of the device~~ The method for introducing molecules as claimed in claim 13, wherein said molecules comprise one of the following: a DNA or a RNA molecule containing regulatory sequences and sequence coding for therapeutic genes or genes of interest for biomedical or biotechnological purposes; an oligonucleotide, (ribo- or deoxyribo-nucleotide, single or double strand, including the SiRNA), whether natural (phosphodiesters) or modified (inside the backbone of the oligonucleotide, ~~such as~~ phosphosulfates, or at the extremities, by addition of groups to protect the oligonucleotides from digestion of nucleases; a protein or peptide, whether natural or genetically or chemically modified, extracted from natural sources or obtained by synthesis, or a molecule simulating the structure of a protein or peptide, whatever its structure; a cytotoxic agent, ~~in particular,~~ the antibiotic bleomycin or the cisplatinum; a penicillin; and other pharmacological agents.